

CLAIMS

1. An autologous culture medium of autologous human progenitor stem cells which comprises:
 - a) between 0.1% and 90% weight of autologous human serum;
 - b) between 0.1% and 10.000 UI/ml heparin;
 - c) between 0.1% and 10.000 UI/ml protamine; and
 - d) a culture medium with basic nutrients with or without glutamine, in sufficient quantity up to 100% weight.
2. A culture medium according to the Claim 1, in which said autologous human serum has been subject to treatments with the aim to inactivate the complement.
3. A culture medium according to the Claim 1, in which said autologous human serum has been obtained from blood samples of the patient.
4. A culture medium according to the Claim 1, in which said autologous human serum has been obtained by the realization of a plasmapheresis to the patient donor of said serum.
5. A culture medium according to the Claim 4, in which said plasmapheresis is carried out using heparin as anticoagulant and protamine sulphate to reverse anticoagulation.
6. A culture medium according to any of the previous Claims which comprises, furthermore, an antibiotic.
7. A culture medium according to Claim 6, in which said antibiotic is selected among penicillin, streptomycin, gentamicin and their mixtures.
8. A culture medium according to any of the previous Claims which comprises, furthermore, amphoterycin B, and/or a Fibroblast Growth Factor (FGF).
9. A culture medium according to Claim 1, which comprises:

89% medium HAM-F12;
10% autologous human serum of the patient
heparin 0.1 at 100UI/ml;
protamine 0.1 at 100UI/ml; y
1% penicillin/streptomycin and, optionally,
0.25mg/ml of amphoterycin B and/or
0.1 at 250 pg/ml of recombinant bFGF.

10. A method for the preparation of an autologous culture medium of autologous human progenitor stem cells, according to any of the Claims 1 to 9, which comprises the mixture of autologous human serum, heparin, protamine, basic nutrients with or without glutamine, together with, optionally, antibiotics, and/or amphoterycin B, and/or a Fibroblast Growth Factor.

11. A method according to Claim 10, in which said autologous human serum has been obtained by plasmapheresis.

12. Use of an autologous culture medium of autologous human progenitor stem cells, according to any of the Claims 1 to 9, for the culture *in vitro*, purification and expansion of autologous progenitor stem cells.

13. A method for the preparation of a composition of autologous human progenitor stem cells, which comprises the incubation of said autologous human progenitor stem cells in an autologous culture medium of autologous human progenitor stem cells, according to any of the Claims 1 to 9 and the purification of the autologous human progenitor stem cells obtained.

14. A method according to Claim 13, in which the purification of the autologous human progenitor stem cells obtained is carried out by the use of specific antibodies which allow the identification of extracellular antigens characteristic of the mentioned autologous human progenitor stem cells.

15. A method according to Claim 14, in which said specific and selective antibodies for the mentioned autologous human progenitor stem cells are joined to magnetic microspheres.

16. A method for the obtention of autologous human muscle progenitor stem cells, useful for their use in cellular therapy, which comprises to incubate said autologous human muscle progenitor stem cells in an autologous culture medium of autologous human progenitor stem cells according to any of the Claims 1 to 9, and purify the autologous human progenitor stem cells obtained.

17. A method according to Claim 16, in which the purification of autologous human muscle progenitor stem cells comprises the use of human anti-CD56 antibodies, optionally, joined to magnetic microspheres, and the selection of the cells which show a phenotype CD56+/ CD45-.

18. A method according to Claim 16, in which the purification of autologous human muscle progenitor stem cells comprises to subject cell culture to a stage of pre-plating with the aim to settle all or part of the fibroblasts present in said cell culture and, subsequently, to identify and separate the autologous human muscle progenitor stem cells by using human anti-CD56 antibodies, optionally, joined to magnetic microspheres, and the selection of the cells which show a phenotype CD56+/ CD45-.

19. A procedure for the obtention of autologous human muscle progenitor stem cells, from a biopsy of muscle tissue, for the preparation of a pharmaceutical composition, which comprises:

a)the realization of a biopsy in a patient object of the subsequent implant of the autologous human muscle progenitor stem cells to extract a fragment of skeletal tissue which comprises autologous human muscle progenitor stem cells;
b)the culture of said autologous human muscle progenitor stem cells from the skeletal muscle in an autologous culture medium of autologous progenitor stem cells according to any of the Claims 1 to 9, under conditions which allow the expansion of said cultured autologous human muscle progenitor stem cells;
c)the purification of said cultured autologous human muscle progenitor stem cells; and
d)the collection of said purified autologous human muscle progenitor stem cells; and optionally,
e)the freezing of said purified autologous human muscle progenitor stem cells until the preparation of said pharmaceutical composition.

20. A procedure according to Claim 19, which comprises the local administration to the patient, in the area of the biopsy, before the carrying out thereof, of a pharmaceutical composition which comprises a pharmacological agent which stimulates the proliferation of autologous human muscle progenitor stem cells.

21. A procedure according to Claim 19, in which said pharmacological agent comprises a local anesthetic, selected between lidocaine and bupivacaine.

22. A method according to Claim 19, in which the purification of autologous human muscle progenitor stem cells comprises to subject the cell culture to a stage of pre-plating with the aim to settle all or part of the fibroblasts present in said cell culture and, subsequently, to identify and separate the autologous human muscle progenitor stem cells by using human anti-CD56 antibodies, optionally, joined to magnetic microspheres, and the selection of the cells which show a phenotype CD56+/ CD45-.

23. Composition enriched in autologous human muscle progenitor stem cells which comprises, at least, 70% of the mentioned autologous human muscle progenitor stem cells in an autologous culture medium of autologous human muscle progenitor stem cells according to any of the Claims 1 to 8.

24. Pharmaceutical composition which comprises, at least, 20 million cells, with a cell density of, at least, 50 million cells/ml and, at least, 40% of autologous progenitor stem cells CD56+/ CD45-, autologous culture medium of autologous progenitor stem cells according to any of the Claims 1 to 9 and, at least, one excipient acceptable from a pharmaceutical point of view.

25. Pharmaceutical composition according to Claim 24, which comprises between 20 and 200 million cells, with a cellular density between 50 and 70 million cells/ml, and at least, 70% autologous progenitor stem cells CD56+/ CD45-, autologous culture medium of autologous progenitor stem cells according to any of the Claims 1 to 9 and human albumin in a quantity between 0,1% and 20% eight with respect to the total quantity.

26. A therapeutical procedure of autologous cellular cardiomyoplasty to create, regenerate and repair dysfunctional myocardial tissue by means of the implant of a pharmaceutical composition which comprises autologous human muscle progenitor stem cells, regenerators of cardiac tissue, and *ex vivo* expanded in an autologous culture medium, comprising said procedure the collection of a material sample from the body of the patient object of the subsequent implant which includes autologous human muscle progenitor stem cells, the expansion of the mentioned cells by culture in an autologous culture medium of autologous progenitor stem cells according to any of the Claims 1 to 9, and the implantation of the collected autologous human progenitor stem cells in the patient, who previously had been extracted such material containing the autologous human muscle progenitor stem cells.

27. A therapeutical procedure of autologous cellular cardiomyoplasty to create, regenerate and repair dysfunctional myocardial tissue by means of the implant of a pharmaceutical composition which comprises autologous human muscle progenitor stem cells, regenerators of cardiac tissue, and *ex vivo* expanded in an autologous culture medium; and where said procedure comprises the following steps:

- a) the collection from the patient of an skeletal muscle biopsy taken from a muscle, preferably, preconditioned by an intramuscular injection of a local anesthetic;
- b) the preparation of a culture medium of autologous human progenitor stem cells, according to any of the Claims 1 to 9, from the autologous serum of the patient;
- c) the preparation of a composition enriched in autologous human muscle progenitor stem cells from the biopsy of a) and the culture medium of b);
- d) the preparation of a pharmaceutical composition from the composition of c); and
- e) the implant of a pharmaceutical composition of autologous human progenitor stem cells of d) in myocardial lesions.

28. A procedure according to Claim 27, in which the implant of said composition of autologous human progenitor stem cells is carried out by direct injection in the peripheral region to the infarction scar or by injection in the intracoronary spaces of both ventriculi.

29. A procedure according to Claim 27, in which the implant of said composition of autologous human progenitor stem cells is carried out by systemic or intracoronary administration by percutaneous venous access.

30. A procedure according to Claim 27, in which the implant of said composition of autologous human progenitor stem cells is carried out by a robotized and computerized system.